Growth and Fecundity of *Lymnaea elodes* (Gastropoda: Lymnaeidae) under Laboratory Conditions

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Abstract. Growth and fecundity of Lymnaea elodes were studied under controlled laboratory conditions. Snails maintained in groups of five showed less significant growth (based on shell length) than snails maintained singly at 5 to 29 weeks after the cultures were initiated. This study used newly hatched juveniles that were maintained in artificial spring water at 22°C and fed leaf lettuce ad libitum. Grouped snails fed either leaf lettuce, Tetramin fish food, or freshly killed Helisoma trivolvis snails (FKS) showed maximal growth based on both shell length and body weight only when maintained on lettuce. Submaximal growth was attained on the FKS diet. Snails maintained on Tetramin grew better than those on FKS, but not as well as those on lettuce. Fecundity studies on mature snails maintained either singly or in groups of five on lettuce showed that egg laying was markedly greater at most data points from snails maintained singly. During the course of this study, only four of about 150 snails died in culture suggesting that this lymnaeid is hardy and well suited for laboratory studies.

INTRODUCTION

Sorensen et al. (1997) identified *Lymnaea elodes* (Say, 1821) (*L. elodes* = *L. palustris* = *Stagnicola palustris* = *S. elodes*) as a vector of the ubiquitous trematode *Echinostoma revolutum* (Froelich, 1802) in the USA. Most studies on growth and fecundity of *L. elodes* are based on observations from natural populations. For example, see the studies of Eisenberg (1966) and Brown et al. (1985). Surprisingly little information is available on growth and fecundity of this snail under controlled laboratory conditions. Because this snail is easy to maintain in the laboratory, achieves a shell length of 2 to 3 cm, and is a vector of *E. revolutum*, it is a useful model for laboratory work.

The purpose of this study was to examine the growth and fecundity of *L. elodes* under controlled laboratory conditions. Specifically, it examined growth and fecundity of snails maintained singly versus in groups. It also examined the effects of different diets on snail growth.

MATERIALS AND METHODS

Lymnaea elodes snails collected from wetlands in northern Indiana, USA (Sorensen et al., 1997) provided the original stocks for this study. Snails derived from these stocks were maintained at 22°C in aerated cultures containing artificial spring water (ASW) prepared as described by Ulmer (1970). Each stock tank contained approximately 50 to 100 snails per 5 L of ASW. Snails were

maintained under diffuse overhead fluorescent light for 12 hr per day and were fed boiled leaf lettuce (*Lactuca sativa*) *ad libitum*. Chalk was added to the cultures as a supplemental source of calcium.

Experiment 1 was done to determine the mean shell length of singly raised snails versus those maintained in a group. A single experiment was done using juvenile snails approximately 1 mm in shell length and was set up with five individual or five grouped snails. To avoid possible genetic factors affecting growth, juveniles were selected from different egg clutches. Single and grouped snails were maintained in 11-cm diameter finger bowls each containing 150 mL of ASW. Snails were fed leaf lettuce ad libitum, and water and food were changed at least twice a week. Snails were measured with a millimeter rule, under a dissecting scope, to the nearest 0.2 mm every week for a total of 29 weeks. During the course of these experiments, in which we examined more than 150 snails ranging from 1 to almost 30 mm in shell length during a span of more than 200 days, mortality was remarkably low in that we had only four deaths.

Experiment 2 determined the effects of various diets on snail shell length and wet weight representing five cultured snails per 11-cm diameter bowl in 150 mL of ASW. This study was initiated using mature snails with a shell length of about 14-mm and a wet weight of about 150 mg. These snails were raised in bowls as described above, and fed *ad libitum* on either leaf lettuce, Tetramin fish food, or freshly killed *Helisoma trivolvis* snails from which the shell was removed and the body severed in half. The mean shell length of *L. elodes* was measured every 5 days for 70 days. Additionally, wet weights of

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these snails were also determined at 5-day intervals for 70 days.

Experiment 3 was done to determine snail fecundity in isolated versus grouped snails, and used sexually mature snails of about 14 mm in shell length. These snails were maintained in bowls as described for Experiments 1 and 2. The total number of eggs was counted, and eggs were removed from the bowls every 5 days up to day 60.

Whenever applicable, the Student's t-test was used to compare differences in means between populations; and a P value of < 0.05 was considered significant.

RESULTS

The results of the study (Experiment 1) on growth of single versus grouped *L. elodes* are presented in Figure 1. Where standard error bars are not shown, they are too small to be included. The shell length increased slowly in both groups until week 4. There was no significant difference (Student's t-test) in shell length in either group up to week 4. Growth became more rapid in both groups between 5 and 21 weeks. Beyond 21 weeks, growth tended to level off in both groups but more so in single snails. From week 5 to 29, growth of single snails was always significantly greater (Student's t-test) than that of grouped snails. Growth differences were most apparent at 20 weeks when shells of single snails were 1.5 times longer in shell length than those of grouped snails.

The results of the study (Experiment 2) on growth of snails maintained on different diets is shown in Figures 2 and 3. As seen in Figure 2, maximal shell length was noted in snails maintained on leaf lettuce. Snails maintained on the freshly killed snail (FKS) diet showed the least increase in size. Snails maintained on the Tetramin diet showed growth intermediate between that of the snails maintained on the other diets. By day 18, the shell length of snails maintained on lettuce was significantly greater (Student's t-test) than that of snails maintained on either Tetramin or FKS. This trend continued for the duration of the experiment. Observations from this experiment using blotted snail wet weights (weights were determined per group of five snails, and averaged to get a weight per snail, precluding the t-test analysis), are shown in Figure 3. Growth here follows the observations seen in Figure 2 in that maximal weight gains were seen in snails fed lettuce, followed by snails fed Tetramin. Least weight gain was seen in snails fed FKS. In fact, in snails fed FKS there was an initial weight loss prior to weight gains beyond day 20.

Fecundity data (Experiment 3) are presented in Figure 4. In general, snails maintained singly laid more eggs than those maintained in groups, and only at one data point (day 5) was this trend reversed. Data showing the greater number of eggs per observation period in single versus grouped snails are shown on days 25 and 45 where the number of eggs laid by snails maintained singly ranged

from about three to eight times greater than that of grouped snails. Duplicate cultures were used when deaths occurred so that measurements on n=5 were always possible. Mortalities never occurred in experiments that used single snails.

DISCUSSION

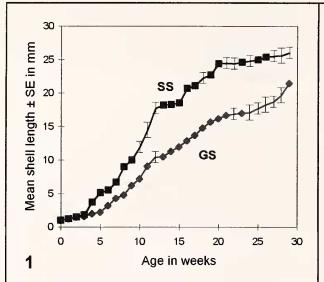
The low mortality observed herein attests to the hardiness of this snail under controlled laboratory conditions using artificial spring water and frequent changes of food and water. These cultures were non-aerated, and bowls were covered with plastic wrap that had been perforated with needles to facilitate gas exchange. The simplicity of our design will be ideal for future use of this lymnaeid in the laboratory for experimentation.

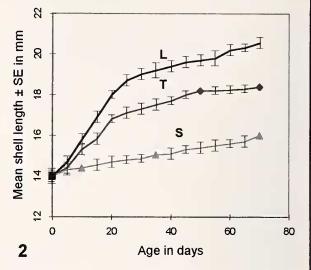
Lymnaeid snails in the laboratory live for approximately 1 year. Data from Hyman (1967) show that lymnaeid longevity varies from 8 to 10 months for *L. palustris*, and up to 15 months for *L. peregra*.

As reviewed by Hyman (1967), many factors influence the growth of pulmonate snails in laboratory cultures including snail density, availability of food, and most importantly, accumulation of waste products. It is not clear why single L. elodes, beginning at about 5 weeks after culture, showed such growth advantages over those maintained as a group of five. The amount of water used and the ad libitum feeding would have supplied more than an adequate water volume and food for this species of snail maintained at five per culture. Perhaps substances released into the water by snails maintained five per group had a growth inhibitory effect on snails of this species. Release of excretory-secretory products by pulmonate snails into water (known as snail-conditioned water or SCW) has multiple effects on conspecific snails, including an influence on intraspecific pairing behavior and attraction of larval trematodes (Marcopoulus & Fried, 1993). Late in the experiment, the grouped snails seemed to be catching up to the single snails in growth. Reasons for this phenomenon are not known.

The effect of diet on lymnaeids has not been thoroughly investigated, although Pennak (1989) noted that some species of *Lymnaea* are omnivorous scavengers, whereas others feed on live animals. In our study, *L. elodes* fed on lettuce, which is suggestive of a herbivorous diet, and fed on Tetramin, which is suggestive of an omnivorous diet. *L. elodes* also fed on freshly killed *Helisoma* snails, which is suggestive of a carnivorous diet. Because the growth of *L. elodes* was optimal on lettuce, the herbivorous diet seems best suited for this lymnaeid, at least under laboratory conditions.

Our observations on fecundity in single versus grouped *L. elodes* did not allow for speculation on the role of self-versus cross-fertilization in this species because snails used in our study came from a sexually mature breeding stock. Therefore, it was not possible to conclude that eggs



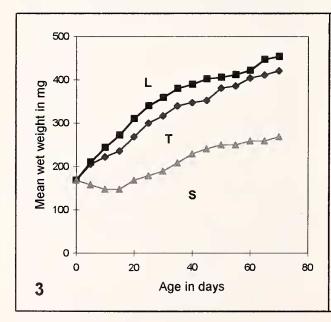


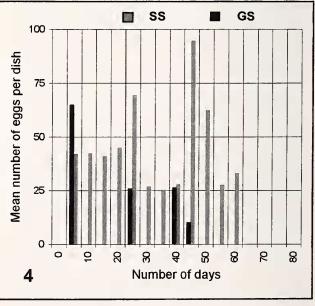
s = Snail diet or FKS diet

T = Tetramin diet

L = Lettuce diet

SS = Single snail
GS = Grouped snail





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laid by our isolated snails resulted from self-fertilization. However, our findings on enhanced egg production in snails maintained singly in *L. elodes* support the observations of van Durvenboden et al. (1985) that *L. stagnalis* maintained singly has a higher oviposition rate than those maintained in groups.

LITERATURE CITED

- Brown, K. M., D. R. De Vries & B. K. Leathers. 1985. Causes of life history variation in the freshwater snail *Lymnaea elodes*. Malacologia 26:191–200.
- EISENBERG, R. M. 1966. The regulation of density in a natural population of the pond snail, *Lymnaea elodes*. Ecology 47: 889–906.
- HYMAN, L. H. 1967. The Invertebrates, Volume VI: Mollusca I. McGraw-Hill: New York. 792 pp.

- MARCOPOULOS, A. & B. FRIED. 1993. Chemoattraction of *Biomphalaria glabrata* (Gastropoda: Planorbidae) for lipid standards and lipophilic factors in leaf lettuce and Tetramin. The Veliger 38:73–74.
- PENNAK, R. W. 1989. Fresh-Water Invertebrates of the United States: Protozoa to Mollusca. 3rd ed. Wiley: New York. 628 pp.
- Sorensen, R. E., I. Kanev, B. Fried & D. J. Minchella. 1997. The occurrence and identification of *Echinostoma revolutum* from North American *Lymnaea elodes* snails. Journal of Parasitolology 83:169–170.
- ULMER, M. J. 1970. Notes on rearing snails in the laboratory. Pp. 143–144 in A. J. MacInnis & M. Voge (eds.), Experiments and Techniques in Parasitology. W. H. Freeman: San Francisco.
- VAN DUIVENBODEN, Y. A., A. W. PIENEMAN & A. TER MATT. 1985. Multiple mating suppresses fecundity in the hermaphrodite freshwater snail *Lymnaea stagnalis:* a laboratory study. Animal Behavior 33:1184–1191.

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Explanation of Figures 1-4

Figure 1. Growth of single (SS) versus grouped (GS) snails. Figure 2. Growth (increase in mean shell length) of snails maintained on various diets. Figure 3. Growth of snails maintained on various diets as described in Figure 2. Growth was based on mean wet weight per snail. Figure 4. Fecundity of single versus grouped snails.